## Corrigendum

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## Corrigendum: Using DNA metabarcoding for assessing chironomid diversity and community change in mosquito controlled temporary wetlands. MBMG 2: e21060. https://doi.org/10.3897/mbmg.2.21060

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## Explanation regarding the background leading to this corrigendum

The main reasons for this corrigendum were discrepancies between the detected number of chironomids in a report from 2014 and the MBMG article, published in 2018. In the MBMG article from 2018, overall more chironomids were reported that were not detected in the report from 2014. The reasons for this are outlined below.

In general, the abundance data used in this study were part of a project conducted in 2013, which investigated the influence of mosquito control with Bti on the food web. This study was approved and financed by the town Neustadt an der Weinstrasse (Germany). The township requested a project report, which was due in March 2014. By this time, the data were not finally evaluated. In particular, a taxonomic training course for determining Chironomidae subfamilies was attended later in the year 2014 by some of the project participants. After this training we found out that many chironomids of the subfamily Orthocladiinae were wrongly determined as Cecidomyiidae. This had direct impacts on the results and created differences in comparison to the report that was published 2014. All detected differences between the report and this manuscript are listed below in Table SI1.

**Table SI1.** Explanation of detected differences in chironomid abundances between the report from 2014 and the manuscript Theissinger et al. (2018), listed by sampling site and sampling week (WAA).

Sampling	WAA	Report 2014	MBMG -	Explanation
site			manuscript	
S	2-13	identical; WAA		This site was not sampled in WAA12.
		12 is missing		
G	2-13	identical; WAA		This site was not sampled in WAA 11. Due to weather conditions sampling in WAA 11 was interrupted, and the few samples
		11 is missing		collected in WAA 11 were pooled with the samples collected in WAA 12.
M	2 –11	identical; WAA		This site was not sampled in WAA12. In this context we detected a typo in Suppl. Material 1 and 5, where we accidently
		12 is missing		indicated WAA12 instead of WAA 13. This mistake has now been corrected. (i.e., sample "12UM" → "13UM" and sample
				"12TM" → sample "13TM"
M	13	~20	~50	On the untreated site M in WAA13 some Orthocladiinae species had been wrongly identified as Cecidomyiidae intead of
				Chironomidae. After instructions of our chironomid specialist and co –author Susanne Michiels, this mistake was corrected.
CL	4	<10	~95	In WAA4 some Orthocladiinae were previously wrongly determined (see above) and only later integrated into the data set.
CL	5	<10	~35	In WAA5 some Orthocladiinae were previously wrongly determined (see above) and only later integrated into the data set.
CL	6	<10	~20	In WAA6 some Orthocladiinae were previously wrongly determined (see above) and only later integrated into the data set.
CL	7	<10	~15	In WAA7 some Orthocladiinae were previously wrongly determined (see above) and only later integrated into the data set.
CL	10	~30	~50	In WAA10 some Orthocladiinae were previously wrongly determined (see above) and only later integrated into the data set.
CL	12	~10	NA	From WAA11 onwards, 5 additional traps were deployed on this site for another side project. In these additional traps 10
				chironomids were collected, which were included in the report, but not later for the manuscript (for comparability reasons).
CL	13	~120	40	This discrepancy is due to the 5 additional traps (see above). The collected chironomids from these 5 traps were included for
				the report, but not in the manuscript.



In this context of clarification, we came across some additional mistakes in the Microsoft Access data base query of the abundance data, where a filter was missing for excluding traps that fell dry during summer. Also, the project in 2013 was conducted over a longer period, namely 18 weeks after initial Bti application (WAA). In the MBMG article we only used the data until WAA13, because after this week too many traps fell dry, with only some data points for aquatic insects remaining. However, in the originally published MBMG article we falsely included emergence data of all individuals until WAA18, including terrestrial taxa from traps that fell dry. The re-calculation of the GLMM still resulted in a 65% significant chironomid reduction in the Bti-treated sites only until WAA4, but not across the entire study period. However, this 65% reduction has to be interpreted with caution because of the strong site heterogeneity and the potentially highly species specific Bti effect (see table below for further details). For full transparency, a new Supplementary Material 3 with all abundance data has now been added to the corrected version of our article.

Due to the strong within-site heterogeneity and the resulting highly variable numbers of chironomids detected per trap within each site, we reduced our overall statement of a clearly negative Bti effect on the chironomid community. In this context we have rearranged and added some text parts, which can be found with tracked changes in the new Supplementary Material 6.

The authors apologize for any inconveniences resulting from these changes and want to explicitly thank the editorial staff for the very thorough review of our study and support in correcting the mistakes.

Finally, we would like to point out that the mistake only affected the overall abundance data, but the metabarcoding part of the study was not concerned and has not been changed.

Below we point out the main changes to be considered by the readers:

Abstract  Methodology – Study sites/ page 3, second paragraph  Methodolgy – Emergence data/ Page 3  Figure 2/ Page 4  Results – Emergence data – 2 <sup>nd</sup> Paragraph/ Page 6  Results – Bti effects on chironomid community composition – 1 <sup>st</sup> paragraph/ Page 7  S  S  S	The re-calculation of the GLMM still resulted in a 65% significant chironomid reduction in the Bti-treated sites only until WAA4, but not across the entire study period. However, this 65% reduction has to be interpreted with caution because of the strong site heterogeneity and the potentially highly species specific Bti effect (see below for further details)  In the last sentence, chironomid community composition "will recover further" has to be changed to "might change".  Further information on discrepancies of Bti treatment on the respective sites has to be considered: The helicopter application took place on April 10, 2013 using IcyPearls (Vectobac WG, ValentBiosciences) at a concentration of 1.44 × 109 TTU/ ha for M and 2.88 × 109 TTU/ ha for S and G. Despite different Bti application rates, we subsequently categorized the further treated sites of the areas M, S and G as "Bti-treated", the first season untreated sites of M, S, G as "untreated" and the control site CL as "never treated". On the application day and over the following week, water depth at all sites varied only marginally (21 to 24 cm).  Further explanation of sampling scheme and timing has to be added: "Samples of traps that fell dry during the sampling period were not evaluated to minimize the presence of terrestrial chironomids in the data set. No sampling was conducted at site G in WAA11 and at sites S, M and CL in WAA12 due to bad weather conditions."  Figure 2 has to be added "No sampling was conducted at site G in WAA11 and at sites S, M and CL in WAA12."  High fluctuation "among traps" have to be highlighted. The term summer emergence peak has to be replaced by "increased emergence"  GLMM output values were corrected for the anaylsis of both time periods, with the total numbers of individuals found at the Bti – treated and Bti –untreated sites until WAA4 added: "The abundance of emergent chironomids until WAA 4 at the Bti-treated sites (N = 162, see Suppl. material 3) was significantly reduced (GLMM t = 9.63, p = 0.01, df = 2) by 65.0%
Abstract  Methodology – Study sites/ page 3, second paragraph  Methodolgy – Emergence data/ Page 3  Figure 2/ Page 4  Results – Emergence data – 2 <sup>nd</sup> paragraph/ Page 6  Results – Bti effects on chironomid community composition – 1 <sup>st</sup> paragraph/ Page 7  S  S  S  S  Results – Study sites/ page 3, Figure 3, Figure 3, Figure 4, Figure 4, Figure 5, Figure 6, Figure 6	the potentially highly species specific Bti effect (see below for further details)  In the last sentence, chironomid community composition "will recover further" has to be changed to "might change".  Further information on discrepancies of Bti treatment on the respective sites has to be considered: The helicopter application took place on April 10, 2013 using IcyPearls (Vectobac WG, ValentBiosciences) at a concentration of 1.44 × 109 ITU/ ha for M and 2.88 × 109 ITU/ ha for S and G. Despite different Bti application rates, we subsequently categorized the further treated sites of the areas M, S and G as "Bti-treated", the first season untreated sites of M, S, G as "untreated" and the control site CL as "never treated". On the application day and over the following week, water depth at all sites varied only marginally (21 to 24 cm).  Further explanation of sampling scheme and timing has to be added: "Samples of traps that fell dry during the sampling period were not evaluated to minimize the presence of terrestrial chironomids in the data set. No sampling was conducted at site G in WAA11 and at sites S, M and CL in WAA12 due to bad weather conditions."  Figure 2 has to be added "No sampling was conducted at site G in WAA11 and at sites S, M and CL in WAA12."  High fluctuation "among traps" have to be highlighted. The term summer emergence peak has to be replaced by "increased emergence"  GLMM output values were corrected for the anaylsis of both time periods, with the total numbers of individuals found at the Bti – treated and Bti –untreated sites until WAA4 added: "The abundance of emergent chironomids until WAA 4 at the Bti-treated sites (N = 162, see Suppl. material 3) was significantly reduced (GLMM t = 9.63, p = 0.01, df = 2) by 65.0% compared to the abundance in the Bti-untreated sites (N = 463; excluding the control site CL; see Suppl. material 3). However, chironomid emergence rates across WAA2 to WAA13 were similar between Bti-treated and untreated sites (GLMM, t = 0.19, p = 0.87, df = 2)."
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	similar between Bti-treated and untreated sites (GLMM, $t = 0.19$ , $p = 0.87$ , $df = 2$ )."
S	
	The first sentence has to be changed to: "Our analysis revealed a negative effect of Bti on chironomid emergence within the first four weeks
	after application." Moreover, it has to be pointed out in more detail that there was "no difference between Bti –treated and Bti –untreated
	sites regarding the chironomid abundance" over the whole study period.
	Species composition and sensitivity of different chironomid species towards Bti has to be discussed in more detail: "Bti toxicity has been
	shown to be highly species specific, not only for mosquitoes but also for chironomids (reviewed in Wolfram & Wenzlund 2018). Our data
	set was comprised of few very dominant and many rare chironomid taxa. On the one hand, some of these rare taxa, such as <i>Limnophyes</i>
	minimus, Dicrotendipes spec., Paratanytarsus spec., Psectrocladius spec. and Tanytarsus spec., are known to be highly Bti sensitive (see
	Wolfram et al. 2018 for a review of studies). More importantly for the results of this study, however, <i>P. uncinatum</i> comprised almost 50% of
	all reads (see Suppl. material 5). Given the strong correlation of read numbers and specimen abundances (Suppl. material 4) <i>P. uncinatum</i>
	is likely the most abundant chironomid taxon in the wetlands studied. If <i>P. uncinatum</i> is a rather Bti –insensitive species, as reported for
	its sister species <i>P. tigrinum</i> (Kondo et al. 1995), this could have overlaid the overall signal of Bti treatment on chironomid abundances. However, broad tests for species specific Bti sensitivity in chironomids are widely lacking (Kästel et al. 2017), making it difficult to make
	overall assumptions of Bti effects on abundance changes."
	The wording on the effect on communities has to be changed to "only prominent" shortly after application
chironomid community composition	The wording on the effect on communities has to be changed to only prominent shortly after application
- 3 <sup>rd</sup> paragraph/Page 10	
Discussion – Bti effects on T	The last sentence highlighting the comparison of four study sites has to be deleted.
chironomid community composition	The last sentence inglinghting the comparison of roar stady sites has to be deleted.
- 4 <sup>th</sup> paragraph/Page 10	
	The wording of the assumption on the chironomid diversity has to be changed to "reduced diversity"
chironomid community composition	The wording of the abbumpton on the elimentative buy has to be changed to reduced according
- 5 <sup>th</sup> paragraph/Page 10	
	The re-calculation of the GLMM still resulted in a 65% significant chironomid reduction in the Bti-treated sites only until WAA4, but not
	across the entire study period. However, this 65% reduction has to be interpreted with caution because of the strong site heterogeneity and
	the potentially highly species specific Bti effect (see above for further details).
	A new Suppl. Material 3 was added showing "All abundance data of aquatic emergence over 13 weeks after Bti application (WAA) across
	all sites (G, M, S, CL), treatment groups (C = Bti –untreated; $T = Bti$ –treated) and traps (1 – 5). This updated Supplemental material 3 also
	includes a new Figure with Chironomid abundances across all traps per site, which have been log transformed for better readability.